

CLAIMS

1. METHOD OF IDENTIFICATION AND QUANTIFICATION OF PROTEINS,
ISOFORMS OF ANGIOTENSIN I CONVERTING ENZYME IN TISSUES,
CELLS AND BIOLOGICAL FLUIDS characterized by the
5 following steps:

- (a) Collecting an aliquot of fresh or concentrated
biological fluids, cells or tissues of living
organisms and submit them to analysis and separation
by Western Blotting method;
- 10 (b) Comparing the sample under analysis to the previous
established standards for the hypertensive genetic
markers and 65 kDa, isoforms of ACE 190 kDa, 90 kDa
and 65 kDa. An aliquot of fluid (for example, fresh
or concentrated urine) using, as analysis control,
15 ACE isoforms prepared as standards and the ACE
recombinant enzyme;
- (c) Detecting the 190 kDa and 65 kDa isoforms presence
in normal individuals and also detecting the
presence of 90 kDa isoforms that is going to
20 characterize those predisposed persons for
developing hypertension and lesions in
characteristic target organs.

2. METHOD according to claim 1, characterized by the fact
that the 90 kDa isoform, which was detected in step (c)

is a hypertension genetic marker and the prognostic agent for hypertension.

3. METHOD according to claim 1 characterized by the fact that chromatographic separation of step (a) is processed
5 AcA44 and/or AcA 34 resin; C-18 reverse phase column C-18, mass spectrometer and Western Blotting using a specific antibody against somatic ACE and against N-domain ACE [90 kDa and 65 kDa] of 190 kDa, 90 kDa and 65 kDa isoforms.
- 10 4. METHOD according to claims 1 to 3 characterized by the fact that the biological fluid is urine.
5. METHOD according to claims 1 to 4 characterized by the fact that it is detected in urine of normotensive individuals, two peaks with angiotensin I converting
15 activity with 190kDa and 65kDa molecular weights.
6. METHOD according to claim 5 characterized by the fact that it is used ion exchange chromatography.
7. METHOD according to claim 5 characterized by the fact that it is detected in hypertensive individual urine a
20 profile where it was eluted two peaks with angiotensin I converting activity with 90 kDa and 65 kDa molecular weights not being detected the 170 kDa form.

8. METHOD OF IDENTIFICATION OF THE POTENTIAL OF 90 KDA ISOFORM OF ANGIOTENSIN I CONVERTING ENZYME characterized by the following steps:

(a) Concentrating and dialyzing the dialyzed urine with Tris-HCl 50 mM buffer, pH 8.0 and submit it to a gel filtration in AcA-34 column equilibrated with Tris-HCl 50mM buffer, containing NaCl 150 mM, pH 8.0;

(b) Collecting 2 mL from the fractions and monitoring them through absorbance measurements at A280nm and by the converting activity of angiotensin I, using Hipuril-L-His-L-Leu- and Z-Phe-His-Leu as subtracts;

(c) Observing the presence of isoforms with ACE activity (170 kDa and 65 kDa) (n=21), from isoforms (170 kDa, 90 kDa and 65 kDa) (n=13) as well as (90 kDa and 65 kDa) (n=13) isoforms.

9. METHOD according to claim 8 characterized by the fact that the two isoforms with ACE activity (170 kDa and 65 kDa) (n=21) detected in the step (c) is from normotensive individuals with normotensive parents.

10. METHOD according to claim 8 characterized by the fact that the three isoforms (170 kDa, 90 kDa e 65 kDa) (n=13) detected in step (c) come from normotensive individuals with hypertensive parents.

11. METHOD according to claim 8 characterized by the fact that the two isoforms (90 kDa and 65 kDa) (n=13)

detected in step (c) come from hypertensive individuals with hypertensive parents.

12. METHOD according to claim 8 characterized by the fact that the 90 kDa isoform is a hypertension genetic marker and a prognostic agent for hypertension.

13. HYPERTENSION GENETIC MOLECULAR MARKER BASED ON SAID GENETIC PROTEINS obtained according to claims 1 to 12 characterized by the fact that it is the basis of the 90 kDa isoform.

14. USE of genetic marker obtained according to claims 1 to 12 characterized by the fact that it is used as a prognostic agent of hypertension.

15. USE of genetic marker obtained according to claims 1 to 12 characterized by the fact that is used in the diagnosis of the predisposition for the development of hypertension and lesions in characteristic target organs.

16. USE according to claim 15 characterized by the fact that the target organs are the heart, nervous system, vascular system and kidney.

17. ANALYTICAL METHOD FOR DIAGNOSIS, RISK STRATIFICATION, THERAPEUTICAL DECISION IN CARRIERS OF ARTERIAL HYPERTENSION AND RENAL LESION characterized by the fact that the presence of de 190 kDa and 65 kDa isoforms are

detected in normal individuals and also it is detected the 90 kDa isoform presence that is going to characterize those predisposed individuals to the development of hypertension and lesions in characteristic target organs.

18. METHOD according to claim 17 characterized by the fact the 90 kDa isoform detected in the step is a genetic marker of hypertension and the prognostic agent of hypertension.

10 19. METHOD according to claim 17 characterized by the fact that the biological fluid is urine.

20. KIT FOR DIAGNOSIS characterized by the fact that it contains the genetic marker obtained according to claims 1 to 12.

15 21. KIT FOR DIAGNOSIS characterized by the fact that it contains the genetic marker and the prognostic agent of hypertension.

20 22. KIT according to claims 16 and 17 characterized by the fact that it is to be used in diagnosis, risk stratification and therapeutical decision in the arterial hypertension.